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Carbohydrate Reserves in Nursery Stock—Effects of Cultural Practices

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ABSTRACT

Planting stock grown at the Lucky Peak Nursery, U.S. Department of Agriculture, Forest Service, near Boise, ID, must be lifted after the winter chilling requirement is satisfied, but early enough in the spring that trees remain dormant. Trees may satisfy their chilling requirement by late fall or early winter and be lifted in December if weather conditions permit. These trees require storage of several months prior to planting. Trees also may be lifted in early spring, but they require storage of several weeks before Intermountain forest planting sites are free of snow. Trees lifted from late fall (November 1) to early spring (March 15) seem better able to maintain their vigor during periods of cold storage than do trees lifted earlier in the fall or later in the spring.

KEYWORDS: total nonstructural carbohydrate, ponderosa pine, lodgepole pine, fertilizer treatment, lift date, moisture stress

When following previously developed procedures for lifting, handling, and planting, survival of planting stock at the Forest Service's Lucky Peak Nursery (near Boise, ID), has generally been successful. However, tree vigor and growth following outplanting are affected by other nursery cultural practices such as irrigation and fertilization regimes. One concern at Lucky Peak is that food reserves necessary for good growth following outplanting are diminished by either long storage time during dormancy or during relatively short storage when trees are not completely dormant. In addition, the effects of various nursery cultural practices on food storage are unknown.

Krueger and Trappe (1967) documented a marked temporal pattern in concentration of total sugars and starch in seedling roots and tops during winter months. They found that sugars increase in concentration starting in November, reach a January peak nearly three times summer concentrations, then decrease steadily to a low level in May. Starch concentrations decrease during the winter sugar buildup, then increase rapidly in March to a mid-April peak. Ronco (1973) studied survival of field planted trees as a function of food reserves but was unable to find a correlation. However, he hypothesized that there must be some threshold level of food reserves required for seedling survival. Van den Driessche (1979) also suggested that a gradual respiratory depletion of stored sugars and starches might be implicated in poor survival and root growth potential. Various published studies have documented that seedling storage invariably results in food reserve depletion: See, for example, McCracken (1979) for starch and sugar declines in mugo and radiata pine through 18 weeks of cold storage, and Ritchie (1982) for descriptions of carbohydrate depletion in Douglas-fir during cold storage.

With the knowledge that storage time will result in depleted food reserves, and the expectation that this may be implicated in poor survival and vigor of planted seedlings, we undertook a study to investigate the effects of various nursery conditions and practices on food storage reserves at time of lifting. Specifically, we tested the hypotheses that nursery soil type, fertilizer treatments, irrigation schedules, lifting dates, and their interactions had an effect on total nonstructural carbohydrate concentration in ponderosa (*Pinus ponderosa* Laws.) and lodgepole pine (*Pinus contorta* Dougl.).

METHODS

We established the study in 1983 on two soils: a granitic sandy loam hereafter referred to as "light soil," and a

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clay loam soil formed from basalt, referred to as "heavy soil." Prior to sowing, both soils had 1 inch (2.5 cm) of sawdust and 400 lb per acre (450 kg per ha) of 34-0-0 incorporated, followed by a 1-year crop of Austrian peas that were plowed under. Soils were fumigated with a mixture of methyl bromide and chloropicrin. Ponderosa and lodgepole pine seeds were sown in standard beds.

We established three watering regimes as follows:

- W1 – Early morning plant water potential < -0.5 MPa during the growing season until the end of irrigation.
- W2 – Plant water potential as above except from July 15 to September 1, when water potential could go to < -1 MPa.
- W3 – As above, except from July 15 to September 1, potential could go to < -1.5 MPa.

Table 1—Fertilizer treatments. For the eight different fertilizer treatments, low N and high N refers to 99 lb per acre and 176 lb per acre (100 and 200 kg per ha) of nitrogen (34-0-0), respectively, incorporated prior to sowing, and three similar applications in the next two growing seasons. P refers to 60 lb per acre (66 kg per ha) of phosphorus (0-46-0) applied once

Fertilizer treatment	Milorganite (presown)		No Milorganite
	1,000 lb/acre	2,000 lb/acre	
1	low N		
2		high N	
3			low N
4			high N
5	low N, P		
6		high N, P	
7			low N, P
8			high N, P

W1 is referred to as the high water regime, W2 as intermediate, and W3 as the low water regime.

Eight fertilizer treatments and a control were assigned to each watering regime. The fertilizer treatments are shown in table 1.

Seedlings were lifted on six dates in fall 1984 and spring 1985. The first lift was September 18 to 21, followed by October 15 to 17, November 5 to 7, November 26 to 28, March 18 to 20, and April 8 to 10. The soil was frozen from mid-December through February, which is common for Lucky Peak Nursery.

Within 4 hours of lifting, five seedlings from each treatment were delivered to the laboratory, washed free of soil, and denatured in a microwave oven to prevent conversion of starches to sugar according to the method of Tiedemann and others (1984). Following the denaturing treatment, trees were oven-dried at 150 °F (65 °C) for 24 hours, then ground in a Wiley mill to pass a 60-mesh screen. Duplicate subsamples of ground tissue were then analyzed for total nonstructural carbohydrate (TNC) by the method of da Silveira and others (1978). TNC includes all carbohydrates fixed by the plant that are generally considered available as food reserves.

Main effects and interactions were determined by analysis of variance for split plots. There were missing values, so covariates were added to the analysis of variance to estimate these values. The design of this study is illustrated in table 2.

RESULTS

Main plot comparisons show significant differences ($\alpha = 0.05$) in TNC concentration between species and soil types and between the three water regimes at $\alpha = 0.10$ (table 2). There were no significant interactions among the three factors—soil, water, and species. Comparison of the

Table 2—Analysis of variance of total nonstructural carbohydrate concentrations by soil types, watering regimes, species, lift dates, and fertilizer treatments

Source	DF	Sum of squares	F value	Pr > F
Main plot comparisons				
Soil	1	34,099.37	34.29	0.0279
Water	2	36,533.39	18.37	.0516
Species	1	24,962.22	25.10	.0376
Soil*Water	2	3,757.87	1.89	.3461
Soil*Species	1	695.23	.70	.4911
Water*Species	2	2,584.47	1.30	.4349
Error A	2	1,988.82		
Subplot comparisons				
Fertilizer	8	8,872.09	2.10	.0510
Soil*Fertilizer	8	4,476.75	1.06	.4045
Water*Fertilizer	16	6,954.62	.82	.6551
Species*Fertilizer	8	4,838.59	1.14	.3487
Error B	56	29,585.14		
Sub-subplot comparisons				
Lift Date	5	1,367,905.34	748.74	.0001
Soil*Lift	5	24,876.99	13.61	.0001
Water*Lift	10	16,960.85	4.64	.0001
Species*Lift	5	18,738.46	10.25	.0001
Fertilizer*Lift	40	14,886.44	1.02	.4129
Error C	464	169,541.38		

means (table 3) indicates that trees grown in the heavy soil had higher concentrations of TNC than trees grown in the lighter soil. These data are presented by lift date in figure 1. There were no significant differences in concentration between soils on the first and third lift dates. Trees grown under high water stress (low water regime) had significantly higher TNC concentration than other trees (table 3). These data are presented in figure 2, plotted by each lift date. Lodgepole pine trees had higher concentrations of TNC than ponderosa pine (table 3, fig. 3). This effect showed up for all lift dates except the first and was significant for all dates combined.

The subplot comparisons for fertilizer treatment indicate a significant main effect at $\alpha = 0.10$. One treatment, 2,000 lb (900 kg) of Milorganite plus subsequent annual high N additions, had a higher mean TNC concentration than did several other treatments including other high N, high N plus P, and one other Milorganite treatment. All four plots treated with Milorganite suffered from pre-emergent damping off disease, and tree densities were lower in these plots. These density differences may have confounded comparisons of fertilizer main effects and fertilizer interactions with other factors. Three of the four Milorganite-treated plots were included in the four highest TNC concentration subplots. Because the significant fertilizer main effect is illogical, we suspect that it is due to some other factor such as tree density.

Table 3—Comparison of mean total nonstructural carbohydrate concentration values for soil types, watering regimes, species, lift dates, and fertilizer treatments. Also included are the 95 percent confidence intervals based on a pooled variance

Treatment	n	TNC, mg/g	Confidence interval
			±
Light soil	324	217.8	3.4
Heavy soil	313	231.5	3.5
High water	214	220.4	4.2
Intermediate water	216	219.3	4.2
Low water	207	234.2	4.3
Ponderosa pine	315	217.4	3.5
Lodgepole pine	322	231.5	3.4
Lift 1 (9/19)	107	155.3	4.4
Lift 2 (10/16)	103	187.8	4.4
Lift 3 (11/6)	107	225.0	4.4
Lift 4 (11/27)	107	243.9	4.4
Lift 5 (3/19)	107	234.6	4.4
Lift 6 (4/9)	106	299.9	4.4
Fertilizer 1	71	225.2	4.5
Fertilizer 2	71	230.4	4.5
Fertilizer 3	66	227.5	4.6
Fertilizer 4	71	221.2	4.5
Fertilizer 5	72	222.1	4.4
Fertilizer 6	72	227.2	4.4
Fertilizer 7	70	223.0	4.5
Fertilizer 8	72	222.5	4.4
Fertilizer 9	72	221.7	4.4

Soil Type vs TNC

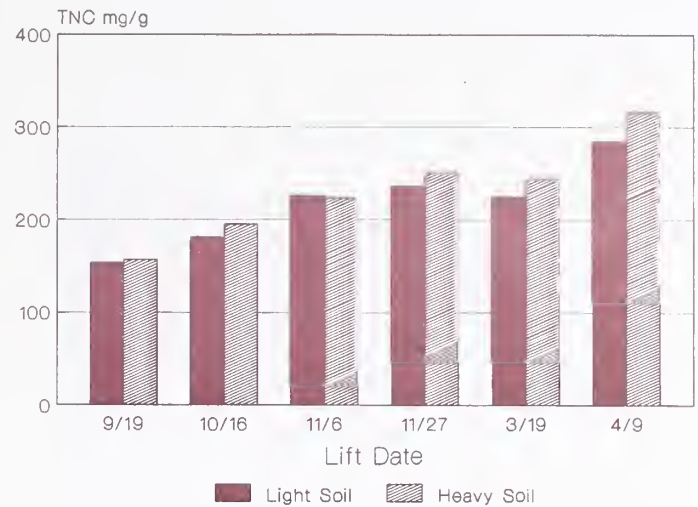


Figure 1—Total nonstructural carbohydrate concentration for seedlings grown in light and heavy soil plotted for each lift date.

Water Regime vs TNC

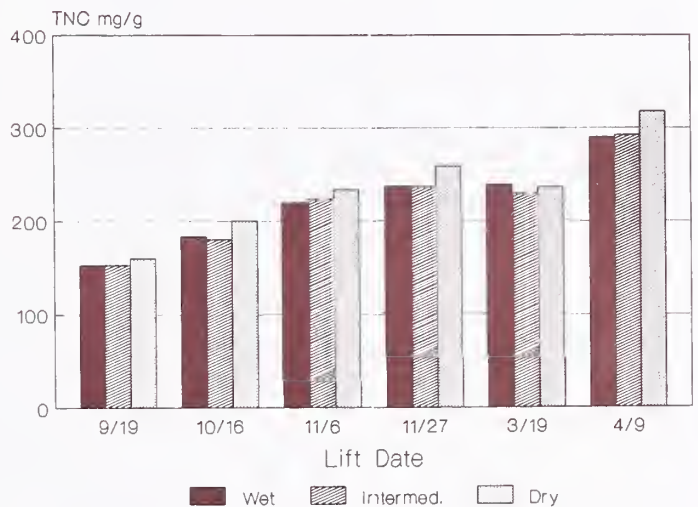


Figure 2—Total nonstructural carbohydrate concentration by water regimes plotted for each lift date.

Species vs TNC

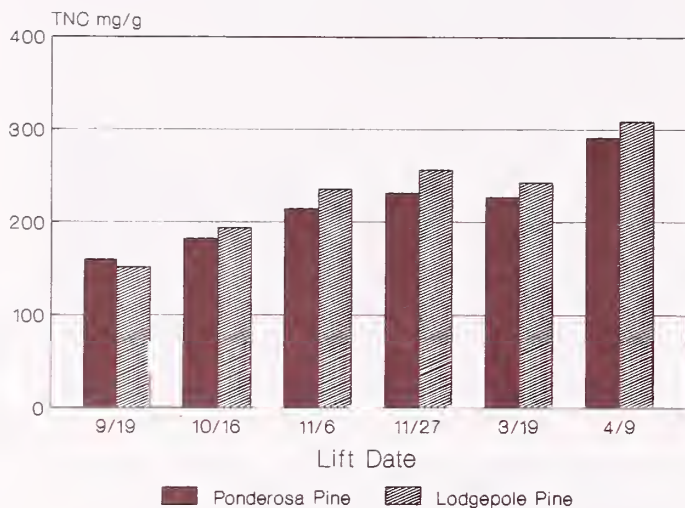


Figure 3—Comparison of total nonstructural carbohydrate concentration by lift date for ponderosa and lodgepole pine.

The sub-subplot comparisons of lift date and lift-date interactions with soil, water, and species all tested highly significant (table 2). Main effects of lift date, plotted on figure 4, show a sharp increase in TNC concentration until early winter, followed by a gradual decline over winter, then an increase in the spring. The strong main effect of lift date, which is exemplified by nearly twice the TNC concentration in lift dates April 8 to 10 compared to lift dates September 18 to 21, assured significant interactions with soil, water, and species.

Lift Date vs TNC

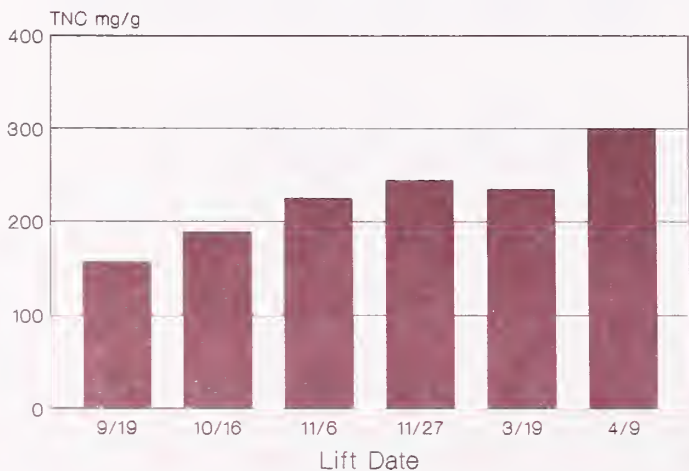


Figure 4—Total nonstructural carbohydrate concentration plotted as a function of lift date.

DISCUSSION

It is not clear why TNC concentrations in trees grown in heavy soil were higher than the concentrations in trees grown in light soil. The heavy soil plots required more frequent watering and more total water added to maintain the plant moisture stress levels. Plant moisture stress levels were often higher in the heavy soil at measurement time, and one might hypothesize that this reduced growth, resulting in smaller trees that reduced shading. This could result in more photosynthate produced per tree and higher photosynthate concentration per tree. However, there were no significant differences in tree size between heavy and light soil. The heavy soil might have held more nutrients. However, the lack of a logical fertilizer effect suggests that N and P were not limiting. It is possible that a higher exchangeable base status in the heavy soil contributed to the TNC difference, but this would require further study to verify.

Trees grown under the low water regime (high moisture stress) had a higher TNC concentration. Trees in the low water regime were significantly shorter, and this could have resulted in less shading and more photosynthate fixed per tree mass, or less respiration of photosynthate per unit time. Either explanation would be consistent with our findings. Iljin (1957) points out that extension growth commonly shows an early response to moisture stress, but photosynthesis continues for some time after growth stops.

Increases in TNC concentrations from lift dates September 18 to 21 to November 26 to 28 reflect low demand for photosynthate relative to production in the late fall, early winter period. This is consistent with Webb's (1977) findings for Douglas-fir seedlings. During the period between lift dates November 26 to 28 and March 18 to 20, when TNC concentration showed a slight decline, snow on the ground covered the seedlings. Following snowmelt, TNC again increased in concentration in the early spring. Photosynthate production probably exceeded demand during this period because of low temperatures. Wardlaw (1968) suggests that growth rates are affected more by low temperatures than photosynthesis, and this is demonstrated by an accumulation of soluble carbohydrate at low temperatures.

CONCLUSIONS

Do the significant differences in TNC concentration as a result of soil type, water regime, and lift date have any implications for nursery practices or planting success? The differences attributable to soil type or water regime were small (approximately 6 percent) and unlikely to affect survival and growth. However, there were large differences in TNC concentration (nearly a factor of two) from the first lift date to early winter or spring. Ronco (1973) found no correlation between survival and food reserves but did hypothesize that there must be some threshold level. This study did not intend to determine a threshold level below which carbohydrates are limiting. Followup studies are needed to determine if there is a threshold level and a field test or a greenhouse-simulated field test to show whether or not the levels found in this experiment make

any difference. Not knowing the relationship between TNC and survival makes it difficult to predict the effect of lift date. However, late fall or spring lifting apparently will result in markedly higher TNC concentrations and may add a measure of safety.

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